Respiratory Medicine (2007) **101**, 1046–1047

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LETTER TO THE EDITOR

Reply to Prof. Bradding and Dr. Brightling

Prof. Bradding and Dr. Brightling have made several remarks concerning our report on mast cell numbers in the airway smooth muscle.[1] With respect to their methodological concerns we would like to respond as follows. First, we regret that in the Materials and methods section, at the end of the paragraph on ''bronchoscopy and processing'', an (essential) sentence is lacking, just before the last sentence. The sentence to be added is (separately sent to the editor as ''corrigendum''): ''Mast cells within and directly in juxtaposition to smooth muscle were counted''. We regret this omission, as this clearly has led to invalid interpretations. The reason we decided to count mast cells not only within but also in direct contact with the smooth muscle is that we wanted to take into account all likely mast cell effects on smooth muscle that could supposedly affect the level of hyperresponsiveness. We realise that we should have made that more clearly in the text.

Further, in addition we actually also did count mast cells within the muscle area separately, with median (range) of 18 (0–53) for asthma biopsies and 17 (0–46) for controls. These numbers are in the same range as the other studies: some report lower,[2] some higher[3–5] numbers. Such differences are likely explained by differences in technique (frozen vs. paraffin vs. plastic-embedded sections) as discussed by Amin et al.[5] Nevertheless, when we performed statistical analysis on mast cell numbers within airway smooth muscle only, this led to the same results between the groups as we reported (no differences; see Table 1). We agree that our findings are different from several other reports, and therefore we have discussed these differences. However, we mainly discussed and pointed out the relation of mast cell numbers to AMP responsiveness. Although we agree that the indicated other studies are of importance (2 of these of the same group),[3,4] these studies did either not have a primary focus on biopsy findings[3,4] or did not include hyperresponsiveness.[3–5]

Our colleagues were mistaken in their suggestion that we only measured two random high power fields. As clearly indicated in the Materials and methods section (third page, first part), we counted all mast cells in two random (whole!) separate sections of a biopsy. The interval between sections was 80 μm, which prevents counting the same cells twice. Further, we also described that we evaluated per subject a mean area of 0.59 mm^2 smooth muscle per subject, which at least equals the measured area in the study of Brightling et al.[2] So this really is similar to the advice to count in- and around the entire airway smooth muscle bundle in a section (in fact we even used two separate sections per biopsy).

So we hope we clarified the issues raised. We believe our results are not contradictory to those of others but in fact an addition to the field, demonstrating that asthma is a heterogeneous disease, in this particular report demonstrating that a difference in bronchial hyperresponsiveness to adenosine-5-monophosphate is not necessarily dependent on differences in mast cell numbers inside and juxtaposed to airway smooth muscle.

References

1. Liesker JJ, Ten Hacken NH, Rutgers SR, Zeinstra-Smith M, Postma DS, Timens W. Mast cell numbers in airway smooth muscle and PC(20)AMP in asthma and COPD. *Respir Med* 2007;**101**(5):882–7.
2. Brightling CE, Bradding P, Symon FA, Holgate ST, Wardlaw AJ, Pavord ID. Mast cell infiltration of airway smooth muscle in asthma. *N Engl J Med* 2002;**346**:1699–705.
3. Berger P, Girodet PO, Begueret H, Ousova O, Perng DW, Marthan R, et al. Tryptase-stimulated human airway smooth muscle cells induce cytokine synthesis and mast cell chemotaxis. *FASEB J* 2003;**17**:2139–41.

Table 1 Mast cell density within airway smooth muscle.

	Control COPD	COPD	Control asthma	Asthma
Number of patients	9	18	9	22
Mast cell density (mast cells/mm^2 airway smooth muscle)	19 (0–155)	13 (0–180)	17 (0–46)	18(0–53)

Data presented as median (range). Mast cell density defined as the number of tryptase-positive spots (mast cells) within smooth muscle, expressed per mm^2 airway smooth muscle area. There are no significant differences between COPD and asthma and their respective age-matched control groups, between asthma and COPD, and between both control groups.

0954-6111/$ - see front matter
doi:10.1016/j.rmed.2007.01.005

4. El-Shazly A, Berger P, Girodet PO, Ousova O, Fayon M, Vernejoux JM, et al. Fraktalkine produced by airway smooth muscle cells contributes to mast cell recruitment in asthma. *J Immunol* 2006;**176**:1860–8.
5. Amin K, Janson C, Boman G, Venge P. The extracellular deposition of mast cell products is increased in hypertrophic airways smooth muscles in allergic asthma but not in nonallergic asthma. *Allergy* 2005;**60**:1241–7.

Jeroen Liesker, Nick ten Hacken, Steven Rutgers, Mieke Zeinstra-Smith, Dirkje Postma, Wim Timens
Departments of Pulmonology and Pathology, University Medical Center Groningen, University of Groningen, The Netherlands
E-mail address: N.H.T.ten.hacken@int.umcg.nl (N. ten Hacken)